Preliminary observations on the comparative gastric morphology of selected Old World and New World bats

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Abstract

Stomachs of ten species of bats (eight microchiropteran and two megachiropteran species), representing nine genera from five families, were examined and compared morphologically and histologically. All stomachs were wholly glandular but differences in gross morphology were observed between insectivorous, omnivorous and frugivorous species. Anatomical variatons in the gastric mucosa between species were minor. The insectivorous bats exhibited little variation between species and possessed a simple tubular gastric stomach. The stomachs of the frugivorous species were more specialised with increased compartmentalisation. Scanning electron microscopy revealed the presence of microvilli in the stomachs of both an insectivorous and a frugivorous species.

Introduction

The gastrointestinal morphology of bats approximates that of other mammals in general organisation (Hill and Smith 1984). Most mammals have a single chambered (unilocular) stomach although ruminants and other foregut fermenters possess a multichambered (plurilocular) stomach which is an adaptation to herbivory (Madge 1975). Feeding styles found among bats include insectivory, piscivory, omnivory, nectarivory, frugivory and sanguinivory, and these are reflected in the anatomy of various regions of the digestive tract.

The gross morphology and ultrastructure of the stomach of many microchiropteran species have been reported (Kamiya and Pirlot 1975; Forman 1972; Rouk and Glass 1970), but little attention has been devoted to the digestive tract of the Megachiroptera (Okon 1977; Bhide 1980; Tedman and Hall 1985a, b). The stomachs of *Epomophorus wahlbergi* and *Eonycteris spelaea* as in *Eidolon helvum* (Okon 1977), *Rousettus leschenaulti* (Bhide 1980), and *Pteropus alecto* and *P. poliocephalus* (Tedman and Hall 1985a, b) are large and saccular with an expanded cardiac and fundic region and a well developed pyloric tube. The pyloric sphincter is asymmetrical. These features are also characteristic of frugivorous microchiropteran species and serve to increase surface area for digestion.

Kamiya and Pirlot (1975) interpreted stomach structure as plesiomorphic in insectivorous bats, transitional in nectar feeders and apomorphic in frugivorous, and especially sanguinivorous, bats. They noted only minor differences in gastric anatomy between Old World and New World insectivorous species and also suggest that the Old World (frugivorous) pteropodids are more specialised for a vegetarian diet than the New World phyllostomatids. It is suggested that the pteropodids followed an independent path of evolution towards vegetarianism before that of the phyllostomatids, and that similarities between the two families are likely a result of convergence (Stephan and Pirlot 1975; Kamiya and Pirlot 1975).

Gastric histology and histochemistry have been used to reveal systematic relationships of bats (Forman 1972; Kamiya and Pirlot 1975), particularly in the Phyllostomatidae.

This examination was undertaken to determine whether gross gastric anatomy can be correlated with feeding specialisations (CARLETON 1973; PERRIN and CURTIS 1979) and phylogeny (PERRIN and CURTIS 1979).

Material and methods

The African species were euthanised with carbon dioxide. Stomachs used for histological and morphological studies were fixed in Bouin's fixative for 18 h after which they were stored in 70 % alcohol. The stomachs of the non-African species, which had been stored in 10 % formalin for up to three years, were also placed in 70 % alcohol. Gross morphology was first examined by bisecting the stomach sagittally. Stomachs less than 1 cm in length were photographed with an Olympus B/H2 photomicroscope and larger stomachs with a Pentax SP II camera fitted with a Pentax macro lens. They were than placed into one of the categories defined by FORMAN (1972). These include: a. "regular", b. "funnelled", and c. "elongated". The authors recognise a further two forms, "recurved" and "tubular" (Table).

Stomachs for histology were embedded in paraffin wax, sectioned at 6–9 μ, and stained with Ehrlich's haematoxylin and eosin, Periodic Acid Schiff (PAS) (HUMASON 1967), or AYOUB and SHKLAR (1983). Light micrographs were taken through a Leitz Laborlux 12 microscope equipped with a Wild Photoautomat MPS 45/51 camera.

For scanning electron microscopy (SEM) small pieces (3 × 3 mm) of stomach tissue were fixed in 3 % cold buffered glutaraldehyde for a minimum of 12 h. The tissue was subsequently critical point dried, coated with gold palladium and viewed with a Hitachi S-570 scanning electron microscope (CROSS 1979).

Results

The terminology used in describing the anatomical form of a hypothetical "generalised" chiropteran stomach is presented in Figure 1.
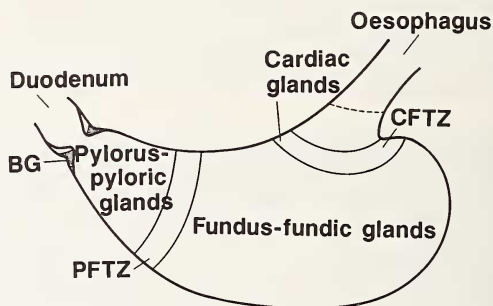

Fig. 1. A diagram to represent a generalised chiropteran stomach. BG = Brunner's glands, CFTZ = cardiofundic transition zone, PFTZ = pyloric-fundic transition zone

Gross morphology

Insectivorous species

The stomachs of the African insectivorous bats, *M. schreibersi, M. fraterculus, R. simulator* and *M. tricolor* are categorised as "regular" and exhibit the simplified tubular gastric form (Fig. 2). They are reniform and generally symmetrical with the greater and lesser curvature nearly parallel. The stomachs are relatively short and the terminal portion (distal to the gastro-oesophageal junction) is equal in length to that of the fornix ventricularis. In *M. tricolor,* however, the fundus is elongated (Fig. 3). The pyloric sphincters of all these species are symmetrical.
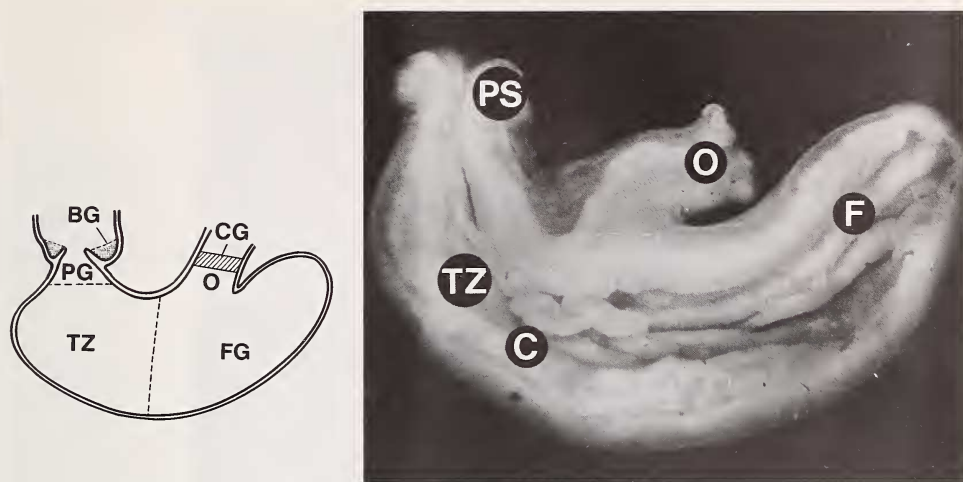
Fig. 2 (left). Diagrammatic representation of a 'regular' chiropteran stomach. BG = Brunner's glands, CG = cardiac glands, FG = fundic glands, PG = pyloric glands, TZ = transition zone. (*Miniopterus schreibersi* × 5,6). – *Fig. 3 (right).* The bisected stomach of *Myotis tricolor* showing the sharply angled oesophagus and elongated fundus. C = corpus, F = fundus, O = oesophagus, PS = pyloric sphincter, TZ = transition zone

Omnivorous species

In the "elongated" stomach of *P. hastatus* the cardiac vestibule is reduced to a vestige (Fig. 4) while in *G. soricina* it is absent (Fig. 5). The fornix ventricularis is dilated and expanded dorsally in both species. The terminal tubular stomach is extensive in *G. soricina*
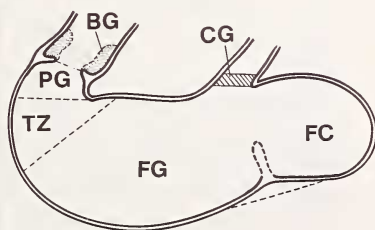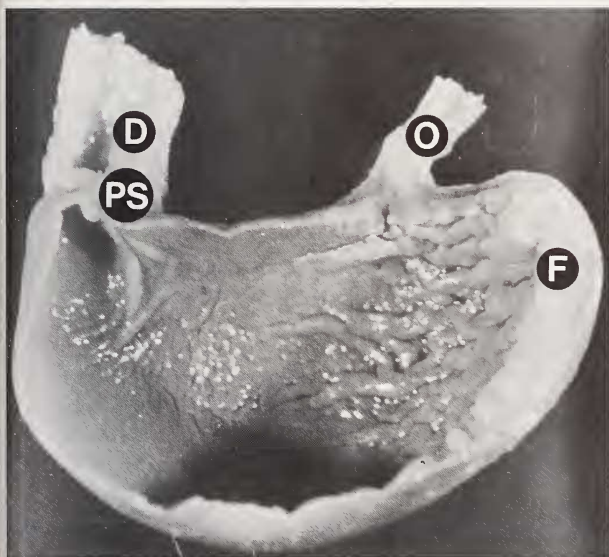

Fig. 4 (left). The bisected stomach of *Phyllostomus hastatus.* D = duodenum, F = fundus, O = oesophagus, PS = pyloric sphincter. – *Fig. 5 (right).* Diagrammatic representation of an 'elongated' chiropteran stomach. BG = Brunner's glands, CG = cardiac glands, FC = fundic caecum, FG = fundic glands, PG = pyloric glands, TZ = transition zone (*Glossophaga soricina* × 6,3)

and *P. hastatus,* and the pyloric sphincters are asymmetrical (since the valves are larger on the greater than the lesser curvature).

Nectarivorous species

Eonycteris spelaea has a markedly sacculated stomach which is extensively elongated, forming a tubular structure (Fig. 6). It is specialised and is here defined as "tubular". The large fornix ventricularis is separated from the fundus by a fold of the stomach wall. The cardiac vestibule is narrow while the fundus forms the elongated tubular portion of the stomach.
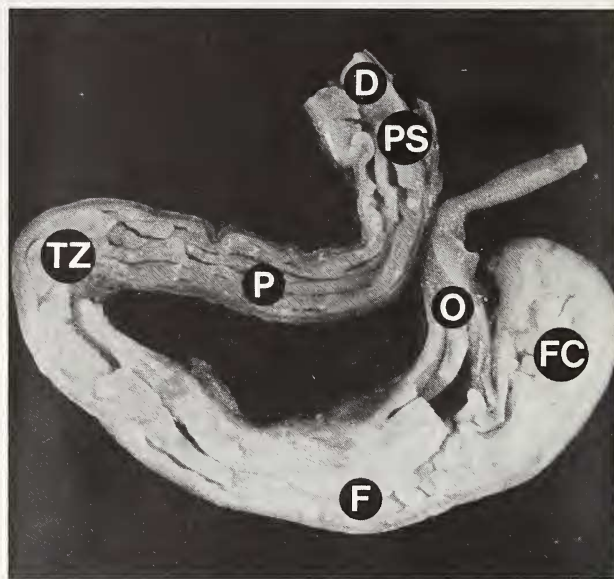

Fig. 6. The bisected stomach of *Eonycteris spelaea.* D = duodenum, F = fundus, FC = fundic caecum, O = oesophagus, P = pylorus, PS = pyloric sphincter, TZ = transition zone

Frugivorous species

Artibeus sp. has a saccular stomach with an unusually large fornix ventricularis and a long broad cardiac vestibule (Fig. 7). The pyloric tube is relatively long and narrow and has a pronounced sulcus intermedius. *Artibeus* exemplifies the "funnelled" stomach condition (Fig. 8).

The stomach of *B. cavernarum* is tubular and reniform and the terminal fundic area is large and recurved (Fig. 9). It is here defined as the "recurved" stomach type. The fundic caecum is also recurved and elongate while the oesophagus enters the stomach at a slight angle. There is a pronounced pyloric constriction and the pyloric sphincter is symmetrical.

E. wahlbergi has a modified 'recurved' saccular stomach with an elongated, recurved fundic caecum (Fig. 10). The fundus, corpus, pyloric antrum and pyloric canal are demarcated externally by constrictions in the stomach wall. The pyloric canal is elongated and the pyloric sphincter is symmetrical.
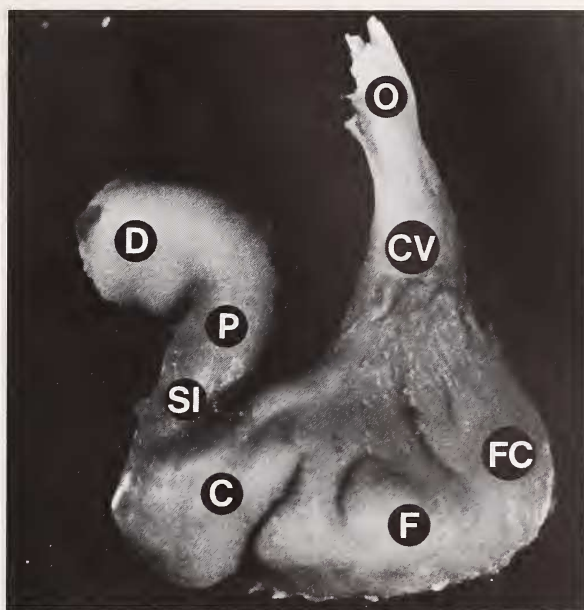
Fig. 7. External view of the whole stomach of *Artibeus* sp. C = corpus, CV = cardia vestibule, D = duodenum, F = fundus, FC = fundic caecum, O = oesophagus, P = pylorus, SI = sulcus intermedius
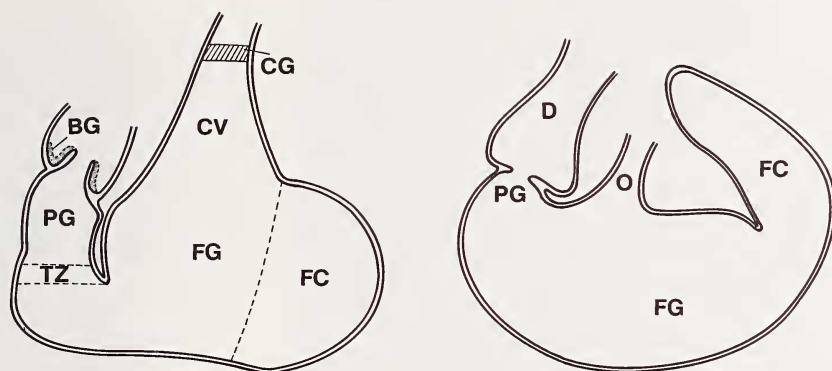

Fig. 8 (left). Diagrammatic representation of a 'funnelled' chiropteran stomach. BG = Brunner's glands, CG = cardiac glands, CV = cardiac vestibule, FC = fundic caecum, FG = fundic glands, PG = pyloric glands, TZ = transition zone. (*Artibeus* sp. × 6,3). – *Fig. 9 (right).* Diagrammatic representation of the 'recurved' stomach condition. FG = fundic glands, PG = pyloric glands, FC = fundic caecum, D = duodenum, (*Brachyphylla cavernarum* × 3,8)

Musculature

The muscularis externa of the insectivorous bats is exceptionally thick throughout the stomach (Fig. 3), and is approximately twice as thick as that of the frugivorous *E. wahlbergi*. In *E. wahlbergi* (Fig. 10), only the circular muscle throughout the greater curvature of the pyloric tube appears to be particularly thickened.
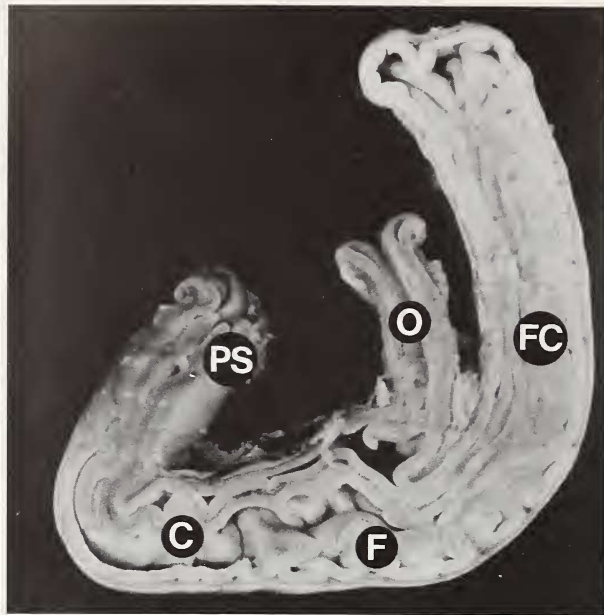
Fig. 10. The bisected stomach of *Epomophorus wahlbergi* (× 3). F = fundus, FC = fundic caecum, O = oesophagus, PS = pyloric sphincter, C = corpus

Pyloric sphincter

The pyloric sphincters of each African insectivorous species examined are symmetrical, and typically possess thick muscle layers. The omnivorous species, *P. hastatus* and *G. soricina*, however, have long asymmetrical valves which are longer on the greater curvature (Figs 4, 5).

In the nectarivore, *E. spelaea*, the musculature is evenly developed on both sides of the pyloric valve. A well-developed valve on the greater curvature is present in *B. cavernarum*. In contrast to the other frugivorous bats, the pyloric sphincter in *E. wahlbergi* is asymmetrical. The musculature is thicker in the valve on the lesser curvature, while the valve on the greater curvature is longer and thinner.

Gastric mucosa

The stomachs of all the species examined are wholly glandular. Fundic glands occupy most of the mucosal surface area, and these are characterised by their long slender appearance, and the presence of chief and parietal cells within the gland (Fig. 11). Under SEM the pyloric and fundic regions are characterised by deep infoldings, the gastric pits. The glands open into these recesses and the columnar epithelial cells show a typical "cobblestone" surface appearance (Fig. 12).

Microvilli are present in the stomach of *M. schreibersi* and are short and sparsely distributed. However, scanning electron microscopy reveals the presence of numerous elongate microvilli throughout the stomach of *E. wahlbergi* (Fig. 13). Yeast and fungal spores (possibly *Penicillium* or *Argostium*) are particularly abundant in the megachiropteran fruit bat, *E. wahlbergi* (Fig. 14), but are also common in the African insectivorous bat species. No bacteria or other symbiotic organisms were observed in any of the bat stomachs examined.

Fig. 11. The fundic glands in the corpus of *Epomorphus wahlbergi* (× 550). Cc = chief cells, GP = gastric pit, Mc = mucoid cells, Pc = parietal cells

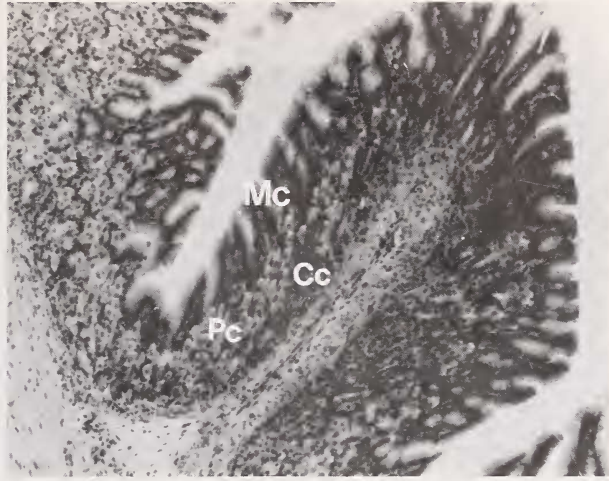*Fig. 12.* A scanning electron micrograph of the "cobblestone" surface of the fundic glands in *Miniopterus schreibersi* (× 480). Ec = epithelial cells, GP = gastric pit
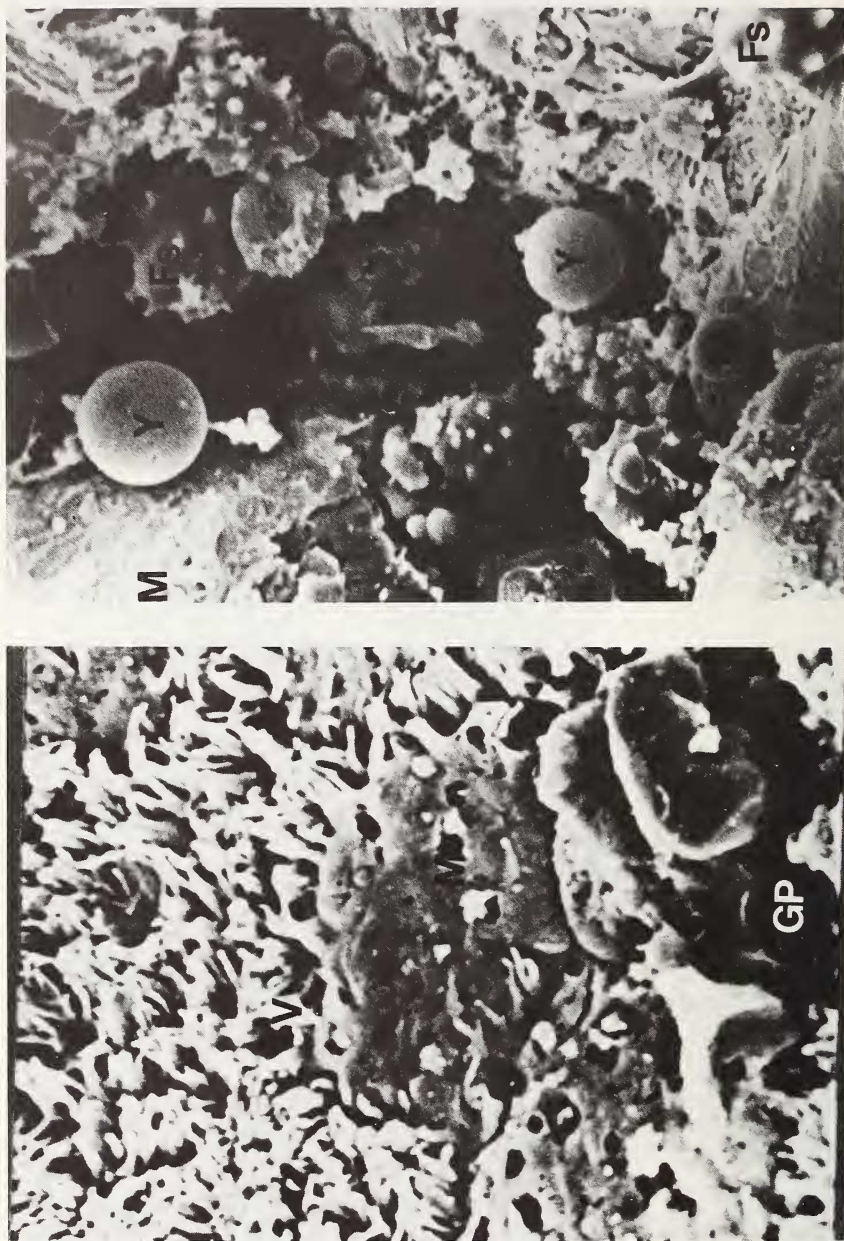
Fig. 13 (left). Scanning electron micrograph of the fundus of *E. wahlbergi*, showing the numerous elongated microvilli. V = microvilli, M = mucus, GP = gastric pit. – *Fig. 14 (right).* Yeast and fungal spores in the stomach of *Epomophorus wahlbergi* (× 3400). FS = fungal spores, M = mucus, Y = yeast cell

Discussion

Gross morphology

Variation in gross gastric morphology is extensive among the bat species examined, but similar trends exist among closely-related genera and among bat species with similar food habits. Several broad categories of gastric anatomy are recognised, named and exemplified. These include regular (*M. schreibersi*), funnelled (*Artibeus* sp.), elongated (*P. hastatus*), recurved (*B. cavernarum*) and tubular (*E. spelaea*).

Characteristics of the bat stomachs studied

Species	Stomach type	Dietary guild	Family
Rhinolophus simulator	Regular	Insectivorous	Rhinolophidae
Myotis tricolor	Regular	Insectivorous	Vespertilionidae
Miniopterus schreibersi	Regular	Insectivorous	Vespertilionidae
Miniopterus fraterculus	Regular	Insectivorous	Vespertilionidae
Epomophorus wahlbergi	Recurved	Frugivorous	Pteropidae
Artibeus sp.	Funnelled	Frugivorous	Phyllostomatidae
Brachyphylla cavernarum	Recurved	Frugivorous	Phyllostomatidae
Phyllostomus hastatus	Elongated	Omnivorous	Phyllostomatidae
Glossophaga soricina	Elongated	Omnivorous	Phyllostomatidae
Econycteris spelaea	Tubular	Nectarivorous	Pteropidae

The funnelled and elongated stomachs have well developed, or reduced/absent, cardiac vestibules respectively, but each has a large fornix ventricularis. The former characterises frugivores while the latter exemplifies a condition intermediate between herbivores and carnivores.

FORMAN (1972) suggests that the sharply-angled pyloric tube of the recurved stomach has a distinctive crook at the point of recurvature, when the stomach is distended. This might serve to retard the rapid transport of food through the stomach and cause foods that are difficult to digest, to be exposed longer to digestive enzymes.

The tubular stomach of *E. spelaea* resembles that of *Artibeus* with respect to the cardiac vestibule and fornix ventricularis. In pteropodids, the cardiac vestibule is more evident than in phyllostomatids (KAMIYA and PIRLOT 1975), while the angled pyloric segment of the pteropodids is as marked as that observed in *Artibeus* sp. Similarities also exist between *E. spelaea* and *Rousettus leschenaulti* (BHIDE 1980), since the stomachs become progressively narrower from the fornix ventricularis to the pyloric region, and the tubular portions reflex sharply.

Musculature

At the gastro-oesophageal junction in *E. wahlbergi*, there is a sphincter-like development of the muscularis externa. This resembles similar structures in *E. helvum* (OKON 1977), *P. alecto* and *P. poliocephalus* (TEDMAN and HALL 1985a, b). It is, however, absent in *R. leschenaulti* (BHIDE 1980). The sphincter may be important in resisting back pressure from liquid food material contained within the stomach, especially when hanging upside down.

The insectivorous bats reveal a much greater degree of muscular hypertrophy throughout the stomach than the frugivorous species. The prominent musculature of the insectivorous bats is of great advantage in increasing the area over which large rhythmical contractions can take place, thereby assisting gastric mixing (FORMAN 1972).

The musculature of the stomach in frugivorous bats is better developed in the fornix ventricularis, cardiac funnel and region of the pyloric sphincter than in the fundus and

corpus. The muscularis externa is, however, less developed than in the insectivorous species. In addition, small grooves are common on the grater curvature, notably in the megachiropteran *E. wahlbergi*. The decrease in thickness of the muscle layer in frugivorous species, and the development of invaginations, is probably an adaptation to allow distension of the stomach to hold large quantities of food.

Pyloric sphincter

Those species possessing a symmetrical pyloric sphincter have a relatively effective closing mechanism which is of greatest advantage to those bats feeding on a diet difficult to assimilate, such as pulpy plants (FORMAN 1972). The frugivorous and nectarivorous species, *B. cavernarum* and *E. spelaea,* possess a symmetrical valve, while the omnivorous species, *P. hastatus* and *G. soricina,* possess a very long asymmetrical valve. The megachiropteran *E. wahlbergi*, however, has an asymmetrical valve similar to that of *R. leschenaulti* (BHIDE 1980).

The African insectivorous bats appear to possess relatively symmetrical valves, and BHIDE (1980) reported that the aperture of the pyloric sphincter in *M. schreibersi* is central. However, many (partially) carnivorous North American bats exhibit some degree of asymmetry of the pyloric sphincter (FORMAN 1972).

Scanning electron microscopy

The presence of microvilli in the stomachs of insectivorous and frugivorous bats has not been reported previously. The microvilli of *E. wahlbergi*, found throughout the stomach, are long, slender and very abundant. The microvilli of the small intestine of *Rousettus aegyptiacus* have been similarly described (KEEGAN and MODINGER 1979). Villi in the small intestine maximise the absorptive surface area (BARRY 1976), and probably account for the rapid digestive process found in megachiropteran fruit bats (KEEGAN and MODINGER 1979; TEDMAN and HALL 1985 a, b). The microvilli in the stomach of *M. schreibersi*, although much shorter and less abundant than in *E. wahlbergi*, likely serve the same function. In the frugivorous bats, gastric microvilli probably increase the surface area for the absorption of carbohydrates, but, in the insectivorous bats, it is likely they function in maximising absorption of glycogen.

The presence of numerous yeast and fungal spores in the stomach of *E. wahlbergi* is not surprising but their occurrence in all the African insectivorous bats examined is intriguing. The bats may ingest spores when feeding on insects, which might themselves have fed on fruit or flowers.

The absence of symbiotic protozoans and bacteria can be explained by the high food transit rate observed in bats (THEDMAN and HALL 1985a) and the fact that herbivory in bats has not been recorded (YALDEN and MORRIS 1975).

Within the family Phyllostomatidae, a gradation exists from insectivorous to omnivorous (*P. hastatus* to *G. spelaea*) to frugivorous habits (*Artibeus* sp.). Dietary differences habe caused a progressive alteration of stomach structure, from the simplified condition in the insectivorous species, to greater elongation and compartmentalisation of the stomach, culminating in the condition found in *Artibeus* sp. This sequence suggests that changes in food habits from the early insectivorous condition resulted in adaptive modifications to stomach morphology.

The uniformity of gross stomach morphology in the four insectivorous species examined in this study lends support to KAMIYA and PIRLOT's (1975) findings of limited variation in gastric structure between insect-eating bats. Variations between frugivorous bats were, however, much greater. The general trend for frugivorous bats, which ingest large quantities of bulky plant material with reduced protein content, is to increase gastric

volume by elongation and sacculation of the stomach, thereby increasing the exposure of food to the digestive processes.

The stomach of the pteropodid *E. spelaea* exhibits the most specialised form of all the bat species examined. This supports Kamiya and Pirlot's (1975) hypothesis that the Pteropodidae followed an independent evolutionary lineage after having diverged from an ancestral stock at a very early stage. The general hypothesis that insectivorous bats constitute the more plesiomorphic taxon (by having a simple tubular stomach), and that the frugivorous species (possessing a complex sacculated gastric structure) represent a more apomorphic taxon, is supported by this study.

Acknowledgements

Zusammenfassung

Vorläufige Befunde zur vergleichenden Magenmorphologie von ausgewählten Altwelt- und Neuwelt-Fledermäusen

Mägen von zehn Fledermausarten (acht Microchiropteren und zwei Megachiropteren) aus neun Gattungen und fünf Familien wurden morphologisch und histologisch untersucht. Alle Mägen waren vollständig mit Drüsenepithel ausgekleidet. Es ließen sich aber morphologische Unterschiede zwischen insektenfressenden, allesfressenden und fruchtfressenden Arten nachweisen. Anatomische Abweichungen der Magenschleimhaut waren unter diesen Arten nur gering. Die insektenfressenden Fledermäuse wiesen geringfügige zwischenartliche Unterschiede auf und hatten einen einfachen röhrenförmigen Magen. Die Mägen der Fruchtfresser waren spezialisierter mit stärker voneinander abgesetzten Abteilungen. Rasterelektronenmikroskopische Untersuchungen wiesen das Auftreten von Microvilli in den Mägen einer insektenfressenden und auch einer fruchtfressenden Art nach.

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